anticipated by, and claims 1, 6, 8-10, 17-23, and 25 as obvious in light of, Bonadio. The Office Action also rejects claims 1, 3, 4, 6, 7, 17, 18, and 22-24 as anticipated by Breitbart. Claims 1, 6, 13, 14, and 18 are rejected as obvious in light of Bonadio and Breitbart; claims 1, 6, 7, 11, 12, 15, 16, and 23 are rejected as obvious over the combination of Bonadio, Breitbart, and Colley; and claims 1 and 5 are rejected as obvious over the combination of Bonadio, Breitbart, and either Ferrara or Neufield.

Discussion of Claim Amendments

Claims 1 and 22 are amended to clarify the identity of the recited angiogenic protein. The subject matter is supported in the Specification, for example, on page 3, lines 1-17.

Claim 4 is amended to clarify the identity of the angiogenic protein. The subject matter is supported in the Specification, for example, on page 3, line 6, and in the Example.

Claim 4 is further amended, and claims 5, 7, and 23 are amended, to further clarify the invention recited therein by removing language drawn to "derivatives." In light of the disclosure in the Specification that the invention includes the use of active derivatives, and given that many active derivatives of the indicated proteins are known, applicants believe that the deletion of the term "derivative" from the claims serves to remove redundant language.

Claims 7 and 23 are amended to clarify the identity of the osteogenic protein by removing recitation of "GMCSF" in light of the recitation of "cytokine" in these claims.

Claim 17 is amended to clarify that the first and second cells are the same cell. This amendment is supported in the Specification, for example, on page 4, lines 10-12.

Claim 18 is amended to clarify that the first and second nucleic acids are the same nucleic acid. This amendment is supported in the Specification, for example, on page 4, lines 35-37 and page 5, lines 14-19.

Claims 23-25 are amended to depend on claim 22, rather than on claim 19.

These amendments add no new matter to the application. For the convenience of the Examiner, a marked-up illustration of the claims as amended, as well as the text of all claims pending upon entry of this Amendment, is attached hereto.

Discussion of Written Description Rejection

The Office Action rejects claims 4, 5, 7, and 23 under 35 U.S.C. § 112, first paragraph, as including matter allegedly not described in the Specification (Office Action, section 1). In particular, the Office Action states that the application does not contain a written description of derivatives of the recited angiogenic and osteogenic proteins. In fact, the Specification makes reference to several published articles disclosing the recited proteins, and active derivatives of

many such proteins are known in the art (see, e.g., Spinella-Jeagle et al., *J. Cell Science, 114*(11), 2085-94 (2001) (abstract enclosed); see also Specification page 3, lines 2-5). However, as discussed above, the claims have been amended to remove the express recitation of the term "derivative," in light of its redundancy. As such, the basis for the rejection for lack of written description is moot, and it should be withdrawn.

Discussion of Enablement Rejection

The Office Action rejects claims 1-25 under 35 U.S.C. § 112, first paragraph, as non-enabled (Office Action, section 2).

As a first basis for the rejection, the Office Action states that the term "region of the bone" is interpreted to mean anywhere in the body; whereas the Specification discloses *in vitro* applications or *in vivo* applications only "in the immediate area of the bone" (Office Action, page 7). Given the language of the Specification, it is urged that even the broadest reasonable interpretation of this term is more narrow than the interpretation set forth in the Office Action. For example, the Specification states that the "region of the bone" includes the bone itself as well as the *immediately adjoining area* within the bone or in tissues surrounding it (Specification, page 2, lines 18-20) – not the entire body. This usage is consistent with the scope of enablement conceded by the Office Action, and it is also consistent with the concept of the "bone progenitor tissue" referenced in the cited Bonadio patent. In conjunction with its concern over the "region of the bone," the Office Action expresses concern over the possible systemic delivery of the reagents for purposes of introducing them into the region of the bone. Applicants note that the Specification discloses methods for *in vivo* topical delivery of the reagents to the desired region of the bone (see, e.g., Specification, page 9, lines 14-24), and it also notes that tropism-altered targeted vectors can be used (Specification, page 7, lines 32-38) as can tissue-specific promoters (Specification, page 5, line 14). It is urged that considerable progress has been made since the articles cited in the Office Action (pages 5-6) on viral targeting, and systemic administration of recombinant adenoviral gene-transfer vectors with bone-specific promoter has been demonstrated to successfully target therapeutic gene expression to cells within bone tissue (see, e.g., Matsubara et al., *Cancer Res., 61*, 6012-19 (2001) (abstract enclosed). It is urged that the Specification, in combination with the state of the art as a whole, enables the delivery of the reagents as recited in the pending claims to the region of a bone, both *in vivo* and *in vitro*.

As a second bases for the rejection, the Office Action asserts that the Specification does not enable the skilled artisan to make "derivatives" of the proteins recited in claims 4, 5, 7, and 23. In fact, it is within the ordinary skill of the art to construct active derivatives of

any of the indicated proteins, and many such derivatives already exist. In any event, the claims have been amended to remove the express recitation of the term "derivative," in light of its redundancy in the context of the application as a whole. As such, the basis for the rejection for lack of written description is moot, and it should be withdrawn.

As a third basis for the rejection, the Office Action asserts that the state of the art would not support an osteogenic role for the hedgehog proteins (Office Action, page 9, citing Lee et al.). The cited Lee et al. paper certainly noted that supplying an Indian hedgehog protein induced chondrogenisis. However, the cartilage that was induced ultimately became ossified, and the article expressly stated that it did not investigate the role of the hedgehog protein in the subsequent osteogenesis. Thus, the Lee et al. article certainly does not rule out a role of hedgehog proteins in osteogenesis. Indeed, other published articles suggest that such proteins can enhance bone density or formation (see, e.g., Spinella-Jaegle et al., enclosed). Moreover, it is known that endogenous BMP promoters respond to Gli proteins (see, e.g., Kawai et al., *Bone, 29*(1), 54-61 (2000) (abstract enclosed)), which are mediators of the hedgehog signal-transduction pathway (*Id.*, see also Lee et al.). Thus, rather than rule out a role of hedgehog proteins in bone formation, the state of the art supports the osteogenic nature of such proteins.

In short, the bases for the rejection for lack of enablement asserted in the Office Action either are misplaced or moot in light of the claim amendments. As such, the rejection under 35 U.S.C. § 112, first paragraph, should be withdrawn.

Discussion of Indefiniteness Rejection

The Office Action rejects claims 1-25 under 35 U.S.C. § 112, second paragraph, as indefinite (Office Action, section 3).

As a first basis for the rejection, the Office Action urges that it is unclear as to what composition is being administered in claim 1 (Office Action, page 10). As amended, claim 1 now recites the identity of this composition. Thus, the basis for the rejection is moot.

As a related basis for the rejection, the Office Action suggests that it is unclear how claim 6 relates to claim 1. Claim 6 specifies that, in addition to the nucleic acid encoding an angiogenic protein, as stated in claim 1, the method also includes the administration of a second nucleic acid encoding an osteogenic protein. The Specification notes that the genes encoding these proteins can be within the same or separate molecules (Specification, page 4, lines 35-37; page 5, lines 14-19). It is urged that one of skill in the art would understand that the respective expression cassettes can be within the same or different nucleic acid molecules. Such alternative arrangements are routinely employed, and the claim is not rendered indefinite because it encompasses either embodiment.

As a second basis for rejection, the Office Action asserts that the term "region of the bone" is indefinite (Office Action, page 11). As noted above, it is urged that those of skill in the art would understand the meaning of this term in the context of the present application.

As a third basis for rejection, the Office Action asserts that the recitation of both "a cytokine" and "GMCSF" in claim 7 renders the claim indefinite (Office Action, pages 11-12). As amended, the claims no longer recite "GMCSF;" hence, the basis for the rejection is moot.

As a fourth basis for rejection, the Office Action suggests that claims 17 and 18 do not clearly indicate the meaning of the first and second cell or amino acid as being the same. This concern is moot in light of the amendments to the claims.

As a final basis for the rejection, the Office Action notes that claims 23-25 contain an incorrect antecedent reference. This concern is moot in light of the amendments to the claims.

In summary, the claims are sufficiently definite to those of skill in the art. As such, the rejection under 35 U.S.C. § 112, second paragraph, should be withdrawn.

Discussion of Anticipation Rejection

The Office Action rejects claims 1-4, 6, 7, 17, 18, 22, and 23 under 35 U.S.C. § 102(e) as anticipated by Bonadio (U.S. Patent 5,942,496) (Office Action, section 4). The Office Action also rejects claims 1, 3, 4, 6, 7, 17, 18, and 22-24 under 35 U.S.C. § 102(e) as anticipated by Breitbart (U.S. Patent 6,077,987) (Office Action, section 5). These two bases for rejection are discussed separately.

A. Bonadio

The Office Action states that Bonadio discloses the transfer of a gene encoding FGF (one of several osteogenic proteins disclosed therein) to cells within a bone progenitor tissue to promote the formation of bone within the tissue. The Office Action also urges that FGF also is angiogenic. Therefore, the disclosure of Bonadio is asserted to anticipate independent claims 1 and 22, as well as claims dependent thereon. As amended, claims 1 and 22 specifies several angiogenic factors that can be employed in the inventive method. However, FGF is not among the factors recited in claim 1, nor does claim 1 include any other factor or protein disclosed or suggested by Bonadio. As such claims 1-4, 6, 7, 17, 18, 22, and 23 are not anticipated by Bonadio.

B. Breitbart

The Office Action states that Breitbart discloses the delivery of vectors having genes encoding FGF or VEGF for enhancing bone growth.

As amended, the independent claims (1 and 22) do not recite the use of FGF. Thus, any disclosure in Breitbart concerning the use of an FGF gene does not disclose the elements of pending claims 1 et seq. nor of claims 22 et seq.

With respect to Breitbart's disclosed use of a gene encoding VEGF, it is urged Breitbart discloses the use of such a nucleic acid to treat skin or wounds. For example, column 3, lines 55-60, identifies VEGF as a "general growth factor important in wound healing". Moreover, column 4, lines 49-54 identifies VEGF as aiding healing, repair, or formation of skin (see also column 6, lines 33-37, listing *VEGF as useful "for wound and skin healing"*), which is disclosed as an alternative to the use of other factors for healing, repair, or formation of bone (column 4, lines 42-45; see also column 6, lines 28-29). Thus, while Breitbart does disclose the use of VEGF for healing skin, the patent does not disclose or suggest the use of VEGF for promoting bone growth. As such, it does not anticipate claims 1, 3, 4, 6, 7, 17, 18, or 22-24.

Discussion of Obviousness Rejection - 35 U.S.C. § 103(a)

The Office Action rejects claims 1, 6, 8-10, 17-23, and 25 as obvious in light of Bonadio (Office Action, section 6). Claims 1, 6, 13, 14, and 18 are rejected as obvious in light of Bonadio and Breitbart (Office Action, section 7). Claims 1, 6, 7, 11, 12, 15, 16, and 23 are rejected as obvious over the combination of Bonadio, Breitbart, and Colley (Office Action, section 8). Claims 1 and 5 are rejected as obvious over the combination of Bonadio, Breitbart, and either Ferrara or Neufield (Office Action, section 9). These bases for rejection are discussed separately.

A. Bonadio – claims 1, 6, 8-10, 17-23, and 25

The Office Action urges that Bonadio discloses the transfer of a gene encoding FGF in combination with genes encoding other osteogenic proteins. It is urged, however, that Bonadio does not disclose or suggest the use of any of the factors of claims 1 or 22, on which the other claims depend. As such, the reference does not suggest all pending claim elements, and it does not render claims 1, 6, 8-10, 17-23, or 25 obvious.

B. Bonadio and Breitbart – claims 1, 6, 13, 14, and 18

The Office Action urges that Bonadio discloses the transfer of a gene encoding FGF in combination with genes encoding other osteogenic proteins, while Breitbart discloses transfecting a cell with a vector encoding VEGF and administering the cells to cranial bone defects to repair the bone defect. As discussed above, Breitbart discloses the use of VEGF only to heal wounds or promote the growth of skin. Indeed, within the disclosure of Breitbart, the use

of VEGF for treating skin is consistently set off as an alternative to using BMP for treating bone. As such, neither Breitbart nor Bonadio discloses or suggests the use of VEGF for enhancing bone density or formation, as recited in claim 1. The combination, thus, does not render obvious claims 1, 6, 13, 14, or 18.

C. Bonadio, Breitbart, and Colley – claims 1, 6, 7, 11, 12, 15, 16, and 23

The Office Action urges that Bonadio discloses the transfer of a gene encoding FGF in combination with genes encoding other osteogenic proteins, while Breitbart discloses transfecting a cell with a vector encoding VEGF to repair bone defects. To this alleged combined teaching, the Office Acton urges that Colley (WO 99/53943) adds the disclosure of using a vector containing a gene encoding midkine or pleiotrophin to enhance bone growth.

As stated above, neither of the primary references discloses the use of a nucleic acid encoding VEGF (or any of the other factors recited in claims 1 and 22) to enhance bone density or formation. Colley does not add to this deficiency of combined teachings. As such, the references do not combine to teach or suggest the subject matter of any of the pending claims. As such, claims 1, 6, 7, 11, 12, 15, 16, and 23 are not obvious in light of the combined teachings of Bonadio, Breitbart, and Colley.

D. Bonadio, Breitbart, and either Ferrara or Neufield – claims 1 and 5

The Office Action urges that Bonadio discloses the transfer of a gene encoding FGF in combination with genes encoding other osteogenic proteins, while Breitbart discloses transfecting a cell with a vector encoding VEGF to repair a bone defect. To this alleged combined disclosure of the primary references, the Office Action asserts that Ferrara et al. (*Endocrine Rev.*, *13*(1), 18-23 (1992)) teach that two derivatives of VEGF (i.e., $VEGF_{121}$ and $VEGF_{165}$) promote angiogenesis. Furthermore, the Office Action asserts that Neufield et al. (*FASEB J.*, *13*(1), 9-22 (1999)) discloses that these same two derivatives induce endothelial cell proliferation and *in vivo* angiogenesis.

While the two secondary references (i.e., Ferrara and Neufield) do disclose the $VEGF_{121}$ and $VEGF_{165}$ derivatives of VEGF, neither of these references suggests using genes encoding any VEGF protein – or any of the other proteins recited in claim 1 - to enhance bone density or formation. As stated above neither of the primary references contains such a teaching either. As such, the references do not collectively disclose the elements of the pending claims. They do not, therefore, combine to render claim 1 or claim 5 obvious.

Conclusion

The application is considered in good and proper form for allowance, and the Examiner is respectfully requested to pass this application to issue. If, in the opinion of the Examiner, a telephone conference would expedite the prosecution of the subject application, the Examiner is invited to call the undersigned attorney.

Respectfully submitted,

Date: August 24, 2001

M. Daniel Hefner, Reg. No. 41826
One of the Attorneys for Applicant(s)
LEYDIG, VOIT & MAYER, LTD.
Two Prudential Plaza, Suite 4900
180 North Stetson
Chicago, Illinois 60601-6780
(312) 616-5600 (telephone)
(312) 616-5700 (facsimile)

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of:

Crystal et al.

Art Unit: 1633

Application No. 09/629,074

Examiner: M. Penn

Filed: July 31, 2000

For: METHOD OF ENHANCING BONE DENSITY

ILLUSTRATION OF AMENDMENTS FILED ON AUGUST 24, 2001

1. (Amended) A method for enhancing bone density or formation, the method comprising administering to at least one first cell associated with a region of a bone at least one first nucleic acid encoding at least one angiogenic protein, such that the first nucleic acid is expressed in the cell to produce the angiogenic protein, whereby bone density or formation is enhanced within the region; wherein the angiogenic protein is a vascular endothelial growth factor (VEGF), a connective tissue growth factor (CTGF), VEGF2, VEGF-C, an angiopoitein, an angiopoetin homologous protein, an angiogenin, an angiogenin-2, or P1GF.

4. (Amended) The method of claim 1, wherein the angiogenic protein is $VEGF_{121}$ or $VEGF_{165}$ [~~a vascular endothelial growth factor (VEGF), a connective tissue growth factor (CTGF), VEGF2, VEGF-C, a fibroblast growth factor (FGF), an angiopoitein, an angiopoetin homologous proteins, an angiogenin, an angiogenin-2, P1GF, or a derivative thereof~~].

5. (Amended) The method of claim 1, wherein the angiogenic protein is selected from the group consisting of $VEGF_{121}$, $VEGFA_{138}$, $VEGFA_{162}$, $VEGF_{165}$, $VEGF_{182}$, and $VEGF_{189}$ [~~,and derivatives thereof~~].

7. (Amended) The method of claim 6, wherein the osteogenic protein is selected from the group consisting of a bone morphogenic protein (BMP), a transforming growth factor (TGF), a latent TGF binding protein (LTBP), latent membrane protein-1 (LMP-1), a heparin-binding neurotrophic factor (HBNF), growth and differentiation factor-5 (GDF-5), a parathyroid hormone (PTH), a fibroblast growth factor (FGF), an epidermal growth factor (EGF), a platelet-derived growth factor (PDGF), an insulin-like growth factor, a growth factor receptor, a cytokine, a chemotactic factor, [~~a granulocyte/macrophage colony stimulating factor (GMCSF)~~] , a LIM mineralization protein (LMP), a leukemia inhibitory factor (LIF), a hedgehog protein, and midkine (MK)[~~, and derivatives thereof~~].

17. (Amended) The method of claim 6, wherein the first cell and the second cell are the same cell.

18. (Amended) The method of claim 6, wherein the first nucleic acid and the second nucleic acid are the same nucleic acid.

22. (Amended) A bone graft comprising at least one first cell having at least one first exogenous nucleic acid encoding at least one angiogenic protein and at least one second cell having at least one second nucleic acid encoding at least one osteogenic protein, wherein the angiogenic protein is a vascular endothelial growth factor (VEGF), a connective tissue growth factor (CTGF), VEGF2, VEGF-C, an angiopoitein, an angiopoetin homologous protein, an angiogenin, an angiogenin-2, or P1GF.

23. (Amended) The bone graft of claim 22[~~19~~], wherein the osteogenic protein is selected from the group consisting of a bone morphogenic protein (BMP), a transforming growth factor (TGF), a latent TGF binding protein (LTBP), latent membrane protein-1 (LMP-1), a heparin-binding neurotrophic factor (HBNF), growth and differentiation factor-5 (GDF-5), a parathyroid hormone (PTH), a fibroblast growth factor (FGF), an epidermal growth factor (EGF), a platelet-derived growth factor (PDGF), an insulin-like growth factor (IGF), a growth factor receptor, a cytokine, a chemotactic factor, [~~a granulocyte/macrophage colony stimulating factor (GMCSF)~~], a LIM mineralization protein (LMP), a leukemia inhibitory factor (LIF), a hedgehog protein, and midkine (MK)[~~.and derivatives thereof~~].

24. (Amended) The bone graft of claim 22[~~19~~], wherein the angiogenic protein is a vascular endothelial growth factor (VEGF).

25. (Amended) The bone graft of claim 22[~~19~~], which is an allograft.

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of:

Crystal et al.

Application No. 09/629,074 Art Unit: 1633

Filed: July 31, 2000 Examiner: M. Penn

For: METHOD OF ENHANCING BONE DENSITY

CLAIMS PENDING UPON ENTRY OF THE AMENDMENT OF AUGUST 24, 2001

1. A method for enhancing bone density or formation, the method comprising administering to at least one first cell associated with a region of a bone at least one first nucleic acid encoding at least one angiogenic protein, such that the first nucleic acid is expressed in the cell to produce the angiogenic protein, whereby bone density or formation is enhanced within the region; wherein the angiogenic protein is a vascular endothelial growth factor (VEGF), a connective tissue growth factor (CTGF), VEGF2, VEGF-C, an angiopoitein, an angiopoetin homologous protein, an angiogenin, an angiogenin-2, or P1GF.

2. The method of claim 1, wherein at least one of the nucleic acids is exposed to at least one cell in vivo in the region of the bone.

3. The method of claim 1, wherein at least one of the nucleic acids is exposed to at least one cell ex vivo, which is then delivered in vivo to the region of the bone.

4. The method of claim 1, wherein the angiogenic protein is $VEGF_{121}$ or $VEGF_{165}$.

5. The method of claim 1, wherein the angiogenic protein is selected from the group consisting of $VEGF_{121}$, $VEGFA_{138}$, $VEGFA_{162}$, $VEGF_{165}$, $VEGF_{182}$, and $VEGF_{189}$.

6. The method of claim 1, further comprising administering to at least one second cell associated with the region at least one second nucleic acid encoding at least one osteogenic protein, such that the second nucleic acid is expressed in the cell to produce the osteogenic protein.

7. The method of claim 6, wherein the osteogenic protein is selected from the group consisting of a bone morphogenic protein (BMP), a transforming growth factor (TGF), a latent TGF binding protein (LTBP), latent membrane protein-1 (LMP-1), a heparin-binding neurotrophic factor (HBNF), growth and differentiation factor-5 (GDF-5), a parathyroid hormone (PTH), a fibroblast growth factor (FGF), an epidermal growth factor (EGF), a platelet-derived growth factor (PDGF), an insulin-like growth factor, a growth factor receptor, a cytokine, a chemotactic factor, a LIM mineralization protein (LMP), a leukemia inhibitory factor (LIF), a hedgehog protein, and midkine (MK).

8. The method of claim 6, wherein the osteogenic protein is selected from the group consisting of BMP-2, BMP-3, BMP-4, BMP-5, BMP-6, BMP-7 and BMP-8.

9. The method of claim 6, wherein the osteogenic protein is TGF-b1.

10. The method of claim 6, wherein the osteogenic protein is BMP-2.

11. The method of claim 6, wherein the osteogenic protein is MK.

12. The method of claim 6, wherein the osteogenic protein is HBNF.

13. The method of claim 6, wherein the angiogenic protein is a VEGF, and the osteogenic protein is TGF-b1.

14. The method of claim 6, wherein the angiogenic protein is a VEGF, and the osteogenic protein is BMP-2.

15. The method of claim 6, wherein the angiogenic protein is a VEGF, and the osteogenic protein is MK.

16. The method of claim 6, wherein the angiogenic protein is a VEGF, and the osteogenic protein is HBNF.

17. The method of claim 6, wherein the first cell and the second cell are the same cell.

18. The method of claim 6, wherein the first nucleic acid and the second nucleic acid are the same nucleic acid.

19. A viral vector comprising at least one first nucleic acid encoding at least one angiogenic protein and at least one second nucleic acid encoding at least one osteogenic protein.

20. The viral vector of claim 19, which is an adenoviral vector.

21. The viral vector 19, which is deficient in at least one essential gene function.

22. A bone graft comprising at least one first cell having at least one first exogenous nucleic acid encoding at least one angiogenic protein and at least one second cell having at least one second nucleic acid encoding at least one osteogenic protein, wherein the angiogenic protein is a vascular endothelial growth factor (VEGF), a connective tissue growth factor (CTGF), VEGF2, VEGF-C, an angiopoitein, an angiopoetin homologous protein, an angiogenin, an angiogenin-2, or P1GF.

23. (Amended) The bone graft of claim 22, wherein the osteogenic protein is selected from the group consisting of a bone morphogenic protein (BMP), a transforming growth factor (TGF), a latent TGF binding protein (LTBP), latent membrane protein-1 (LMP-1), a heparin-binding neurotrophic factor (HBNF), growth and differentiation factor-5 (GDF-5), a parathyroid hormone (PTH), a fibroblast growth factor (FGF), an epidermal growth factor (EGF), a platelet-derived growth factor (PDGF), an insulin-like growth factor (IGF), a growth factor receptor, a cytokine, a chemotactic factor, a LIM mineralization protein (LMP), a leukemia inhibitory factor (LIF), a hedgehog protein, and midkine (MK).

24. The bone graft of claim 22, wherein the angiogenic protein is a vascular endothelial growth factor (VEGF).

25. The bone graft of claim 22, which is an allograft.